

Western Blot of Flk-1/SEAPS Immunoprecipitation with MAb DC101

FIK-1 AP

FIGURE 2A

Probe: Anti-Ptyr



FIGURE 2B

MAb (μg/ml) - - .5 1 2.5 5 VEGF (40 ng/ml) - + + + + + - 200

Probe: Anti-Ptyr



FIGURE 3A

MAb (5μg/ml) VEGF (ng/ml)

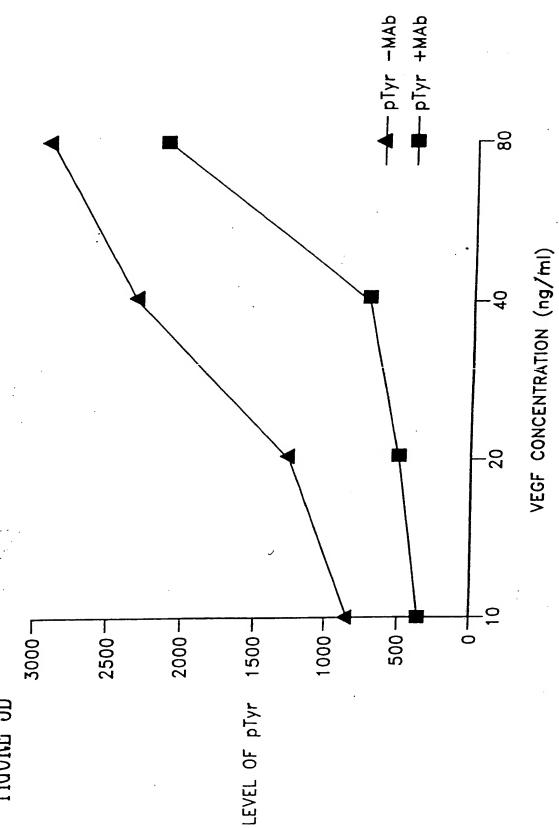
80 40 20 10 80 40 20 10

-

200

Anti-Ptyr







Inhibition of VEGF-Flk-1/fms activation by pre-bound MAb DC101

Assay conditions:

PC

41.086.31

MAb (5 μ g/ml):

· + - - 🛧 👍

VEGF (ng/ml):

- 20 40 40 40



Probe: Anti-Ptyr

Assay conditions: P: MAb pre-bound 15'; VEGF 15'

C: Competitive assay; MAb

+ VEGF 15'



- 10 2.5 10 SE CM 20 40 20 40 40/ MAb (μg/ml) - 2.5 10 VEGF₁₆₅ (ng/ml) - - -

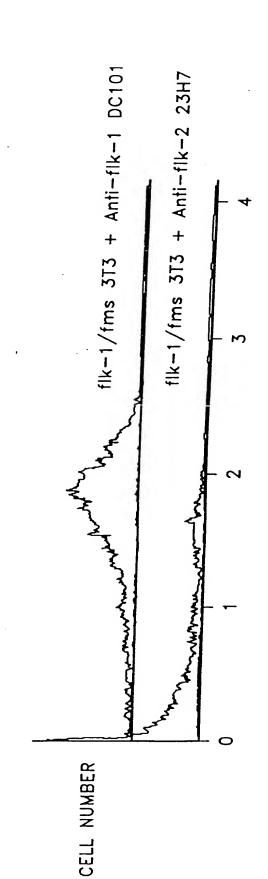


- 200

Probe: Anti-Ptyr

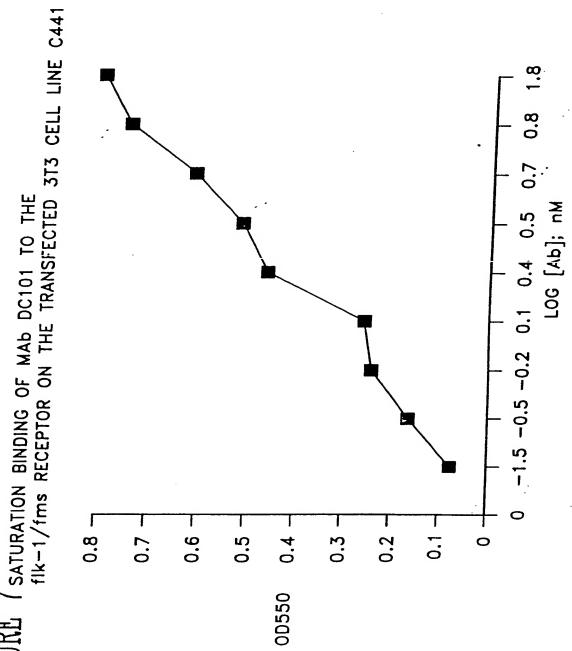
- 97

FIGURE 6 facs analysis of anti-fik-1 mad binding to fik-1/fms transfected 373 cells (C441)



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Immunoprecipitation of phosphorylated Flk-1/fms from VEGF stimulated Flk-1/fms transfected 3T3 cells

Antibody: 1 2 3 4

- 200

Anti-pTyr

Antibodies:

- 1) Rat anti-flk-2 lgG_{2a} 2A13
- 2) Rat anti-flk-1 IgG₁ DC101
- 3) Rat anti-flk-2 IgG₁ 23H7
- 4) Rabbit anti-fms polyclonal IM 133



Sensitivity of VEGF induced phosphorylation of the Flk-1/fms receptor to inhibition by MAb DC101

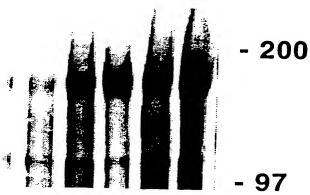
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Effect of MAb DC101 on CSF-1 induced phosphorylation of the fms receptor

MAb (5μg/ml) CSF-1 (ng/ml)

- + - - + + - - 20 40 20 40



Probe: Anti-Ptyr



Specificity of MAb DC101 neutralization of the activated Flk-1/fms receptor

Condition: VEGF ng/ml: MAb: (5μg/ml)

Competitive 20 40 40 40 40 Prebound 40 40 40 2 3





- 200

Anti pTyr





Anti-FMS

Rat MAbs: 1) Anti-fik-1 IgG₁ DC101 2) Anti-fik-2 IgG₁ 23H7 3) Anti-fik-2 IgG_{2a} 2A13

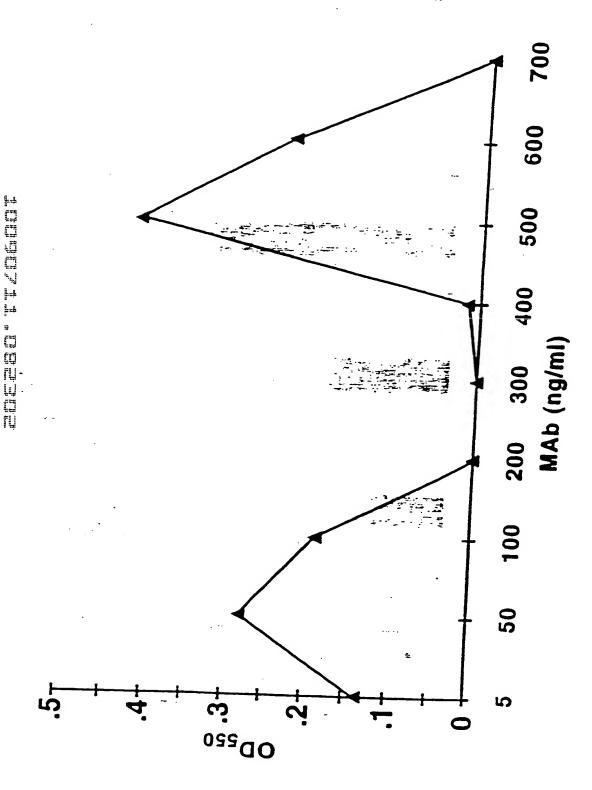


Figure 12

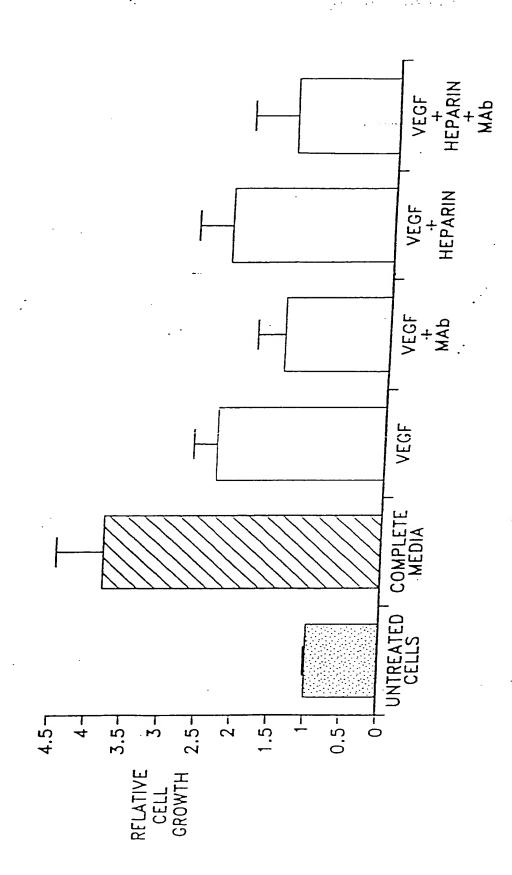


Immunoprecipitation of phosphorylated receptor bands from VEGF stimulated HUVEC cells

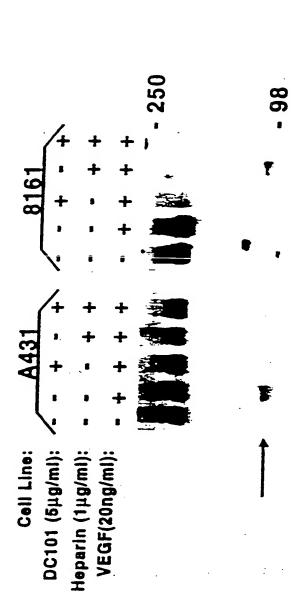
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FIGURE 14





Effect of MAb DC101 on VEGF Receptor Forms in the Tumor Cell Lines A431 and 8161



IP: MAb DC101 Probe: Anti-pTyr

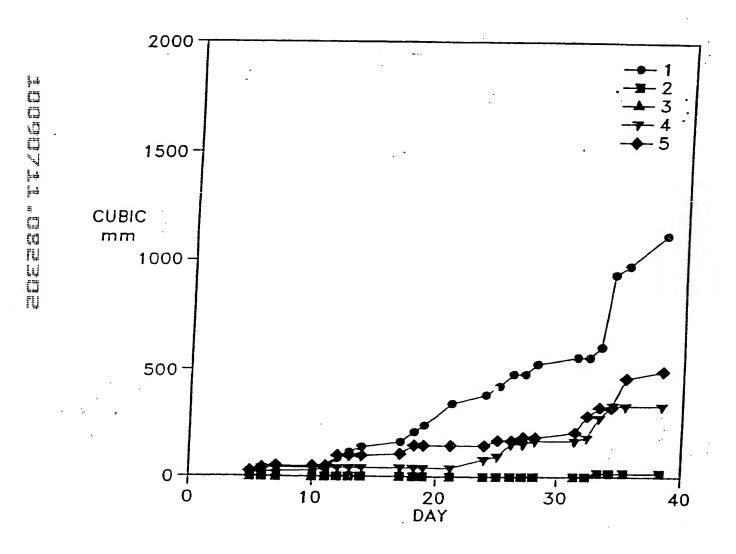
Figure 15

Ante 12



FIGURE 16A

TREAMENT OF GLIOBLASTOMA
XENOGRAFTS WITH RAT anti-flk-1 MAb



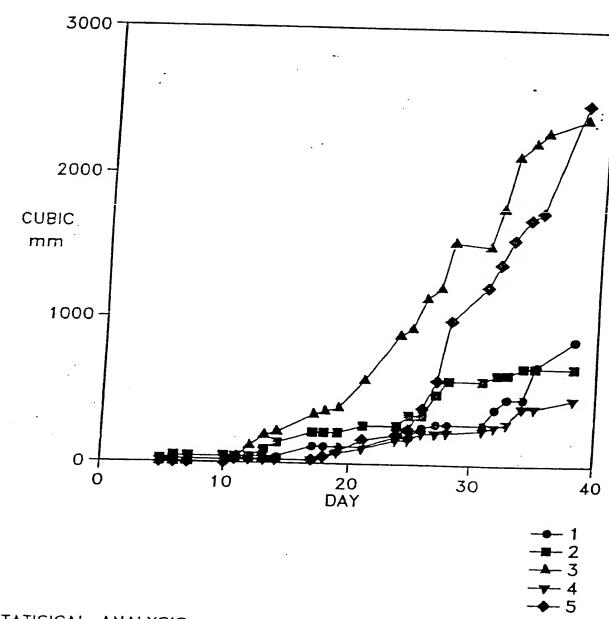
STATISICAL ANALYSIS flk-1 SLOPE = 16.09 p VALUE FOR flk-1 VERSUS flk-2 TUMOR SIZE = 0.0001

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FIGURE 16B

TREAMENT OF GLIOBLASTOMA XENOGRAFTS WITH RAT anti-flk-2 MAb



STATISICAL ANALYSIS
flk-2 SLOPE = 37.39
p VALUE FOR flk-1 VERSUS flk-2 TUMOR SIZE = 0.0001



